

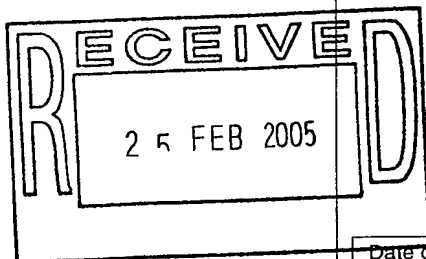
PATENT COOPERATION TREATY

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

PCT

To:

NASH, Keith Wilfrid
KEITH W NASH & CO.
90-92 Regent Street
Cambridge CB2 1DP
GRANDE BRETAGNE



NOTIFICATION OF TRANSMITTAL OF
THE INTERNATIONAL PRELIMINARY
EXAMINATION REPORT
(PCT Rule 71.1)

Date of mailing
(day/month/year)

24.02.2005

Applicant's or agent's file reference
C1435.01/P

IMPORTANT NOTIFICATION

International application No.
PCT/GB 03/04486

International filing date (day/month/year)
15.10.2003

Priority date (day/month/year)
16.10.2002

Applicant
PERKINELMER UK LIMITED et al.

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.
2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.
3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translation to those Offices.
4. **REMINDER**

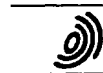
The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices) (Article 39(1)) (see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the PCT Applicant's Guide.

The applicant's attention is drawn to Article 33(5), which provides that the criteria of novelty, inventive step and industrial applicability described in Article 33(2) to (4) merely serve the purposes of international preliminary examination and that "any Contracting State may apply additional or different criteria for the purposes of deciding whether, in that State, the claimed inventions is patentable or not" (see also Article 27(5)). Such additional criteria may relate, for example, to exemptions from patentability, requirements for enabling disclosure, clarity and support for the claims.

Name and mailing address of the international
preliminary examining authority:



European Patent Office - P.B. 5818 Patentlaan 2
NL-2280 HV Rijswijk - Pays Bas
Tel. +31 70 340 - 2040 Tx: 31 651 epo nl
Fax: +31 70 340 - 3016

Authorized Officer

Wach, P

Tel. +31 70 340-3325



PATENT COOPERATION TREATY
PCT
INTERNATIONAL PRELIMINARY EXAMINATION REPORT
(PCT Article 36 and Rule 70)



Applicant's or agent's file reference C1435.01/P	FOR FURTHER ACTION		See Notification of Transmittal of International Preliminary Examination Report (Form PCT/PEA/416)
International application No. PCT/GB 03/04486	International filing date (<i>day/month/year</i>) 15.10.2003	Priority date (<i>day/month/year</i>) 16.10.2002	
International Patent Classification (IPC) or both national classification and IPC G02B21/00			
Applicant PERKINELMER UK LIMITED et al.			

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 6 sheets, including this cover sheet.
 - ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

These annexes consist of a total of 6 sheets.

3. This report contains indications relating to the following items:
 - I ☒ Basis of the opinion
 - II ☐ Priority
 - III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
 - IV ☐ Lack of unity of invention
 - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
 - VI ☐ Certain documents cited
 - VII ☐ Certain defects in the international application
 - VIII ☐ Certain observations on the international application

Date of submission of the demand 08.05.2004	Date of completion of this report 24.02.2005
Name and mailing address of the international preliminary examining authority:  <div style="margin-left: 20px;"> European Patent Office - P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk - Pays Bas Tel. +31 70 340 - 2040 Tx: 31 651 epo nl Fax: +31 70 340 - 3016 </div>	Authorized Officer Ward, S Telephone No. +31 70 340-3547 

**INTERNATIONAL PRELIMINARY
EXAMINATION REPORT**

International application No. PCT/GB 03/04486

I. Basis of the report

1. With regard to the **elements** of the international application (*Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17)*):

Description, Pages

1-34 as originally filed

Claims, Numbers

1-31 filed with telefax on 27.01.2005

Drawings, Sheets

1/15-15/15 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
☐ the language of publication of the international application (under Rule 48.3(b)).
☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
☐ filed together with the international application in computer readable form.
☐ furnished subsequently to this Authority in written form.
☐ furnished subsequently to this Authority in computer readable form.
☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- ☐ the description, pages:
☒ the claims, Nos.: 32-172
☐ the drawings, sheets:

**INTERNATIONAL PRELIMINARY
EXAMINATION REPORT**

International application No.

PCT/GB 03/04486

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

☐ the entire international application,

☒ claims Nos. 2-15, 17-31

because:

☐ the said international application, or the said claims Nos. relate to the following subject matter which does not require an international preliminary examination (specify):

☐ the description, claims or drawings (*indicate particular elements below*) or said claims Nos. are so unclear that no meaningful opinion could be formed (*specify*):

☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

☒ no international search report has been established for the said claims Nos. 2-15, 17-31

2. A meaningful international preliminary examination cannot be carried out due to the failure of the nucleotide and/or amino acid sequence listing to comply with the standard provided for in Annex C of the Administrative Instructions:

☐ the written form has not been furnished or does not comply with the Standard.

☐ the computer readable form has not been furnished or does not comply with the Standard.

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes: Claims	1,16
	No: Claims	
Inventive step (IS)	Yes: Claims	
	No: Claims	1,16
Industrial applicability (IA)	Yes: Claims	1,16
	No: Claims	

2. Citations and explanations

see separate sheet

Re Item V

Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. For the reasons mentioned under item III, the reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability is limited to claims 1 and 16 (based on claims 1 and 97 as originally filed).
2. **Reference is made to the following documents:**
D2: US20020097490 A
D3: US4566029 A
D4: US4910606 A
D5: JP2266674 A
- 3.1 The subject-matter of claim 1 does not involve an inventive step in the sense of Article 33(3) PCT. The document D2 discloses (see e.g. figure 19): A method of imaging light from a specimen (40) in which excitation light passes to the specimen via a confocal scanning system and light emitted by luminescence (see paragraphs [0221], [0222]) of the specimen passes in the other direction via the scanning system to an image capture device (46) having a sensor having discrete spatially distinct light sensitive regions (a CCD camera, see paragraph [0221]), and the scanning system is operated so as to scan the whole of an area of interest of the specimen, wherein the scanning system and the image capture device are controlled by a controller (86,78), so that the light emitted from the specimen is incident on the image capture device for a specific time period equal to that required for scanning the whole of the area of interest n times, where n is a whole number equal to or greater than 1 (i.e. the time periods determined by sectors 102a, 102b etc. in fig 20).
- 3.2 A further feature of claim 1 is that "the scanning system, and the excitation light and/or the image capture device are controlled by **a controller programmed to act as a state machine**". The term "state machine" refers to a very general model in computer science which may be implemented in many ways. The application gives one detailed implementation of a state machine (page 26, final paragraph, pages 27-32 and fig 4). However, many other implementations are possible, and in particular it is well known that a general purpose digital computer can be regarded as satisfying all the defining criteria of a "state machine", and therefore one possible implementation of a "state machine" is a

computer, e.g. computer (18) in figure 19 of D2. Hence in D2 the scanning system and the image capture device are controlled by a controller which is programmed to act as a state machine.

- 3.3 Claim 1 therefore differs from D2 in that the scanning system and the excitation light and/or the image capture device are controlled by a controller, so that the light emitted from the specimen is only incident on the image capture device for a specific time period equal to that required for scanning the whole of the area of interest n times, where n is a whole number equal to or greater than 1.
- 3.4 In D2, outside the scanning periods relatively little light will be reflected from the disk to the image capture device (as a result of the black sectors, e.g. 102d, 102e, 102f in fig 20), but other light (e.g. stray light coming directly from the source (30) in fig 19) may still be incident on the image capture device even in the non-scanning periods.
- 3.5 It is commonly known in the field of video imaging that solid state image capture devices, such as the CCDs of D2, suffer from a problem referred to as "smear", "dark charge" or "dark current", whereby during the charge transfer period when the camera should not be performing an exposure operation, light incident on the device nevertheless results in some extra unwanted charge being produced which is detrimental to the image. The problem associated with claim 1 may therefore be seen as reducing image errors (see the description of the present application, page 6, paragraph 2).
- 3.6 A solution to this problem is also well known in the art, namely to allow light to be incident on the image capture device only in the exposure periods, and to prevent light from being incident on the image capture device in the non-exposure periods. In implementing such a solution in the context of D2, it would be obvious for the skilled person to use the control arrangements (78,86) to apply the appropriate synchronization signals. Hence, starting from D2 and applying this commonly known solution to overcome the above-mentioned problem of "smear", the person skilled in the art would arrive, in an obvious manner, at the features of claim 1. Hence claim 1 lacks inventive step (Article 33(3) PCT).
- 3.7 For purposes of illustration, the following documents provide examples of overcoming the problem of "smear" by preventing light from being incident on the image capture device in the non-exposure periods:

**INTERNATIONAL PRELIMINARY
EXAMINATION REPORT - SEPARATE SHEET**

International application No. PCT/GB 03/04486

- D3: uses rotary chopper (see abstract, figures).
 - D4 uses LC, PLZT, mechanical or rotary shutter (see column 6, lines 28-33)
 - D5 uses LC shutter (see abstract, figures).
4. The features of Independent apparatus claim 16 correspond to those of method claim 1, and hence claim 16 does not involve an inventive step in the sense of Article 33(3) PCT for the reasons given above, *mutatis mutandis*.
5. Furthermore, it is pointed out that claim 1 implies that light is only incident on the image capture device for "a specific time period". In the description it appears that light is incident on the image capture device during several time periods corresponding to periodic exposures (see e.g fig 6a). This contradiction between the claims and description is contrary to Article 6 PCT.

01.02.2005

(71)

C1436.01/P

CLAIMS

1. A method of imaging light from a specimen in which excitation light passes to the specimen via a confocal scanning system and light emitted by luminescence of the specimen passes in another direction via the scanning system to an image capture device having a sensor having discrete spatially distinct light sensitive regions, and the scanning system is operated so as to scan the whole of an area of interest of the specimen, wherein the scanning system, and the excitation light and/or the image capture device are controlled by a controller programmed to function as a state machine so that light emitted from the specimen is only incident on the image capture device sensor for a specific time period equal to that required for scanning the whole of the area of interest n times (where n is a whole number equal to or greater than 1).
2. A method as claimed in claim 1 wherein shutter means is provided which operates to prevent light reaching at least part of the image capture device sensor, except for the said specific periods of time during which the excitation light is incident on the specimen, for the purpose of reducing errors which can arise from light arising from phosphorescence, afterglow, stray reflections or other effects, from reaching the capture device sensor.
3. A method as claimed in claim 1 or claim 2 wherein the specimen is at least in part transparent and a plurality of images are formed by scanning the specimen in a plurality of different spaced apart planes.
4. A method as claimed in claim 3 wherein the different planes are produced by relative movement between the specimen and a scanning device forming part of the scanning system.

5. A method as claimed in claim 3 wherein the different planes are produced by movement of at least one part of an optical system forming part of the scanning system so that light is brought to a focus in the specimen at different spaced apart points, each point therefore defining the position of a focal plane of the scanning system.
6. A method as claimed in claim 4 or claim 5 wherein movement is restricted to periods during which excitation light is not incident on the specimen.
7. A method as claimed in claim 4 or claim 5 wherein movement is restricted to periods during which the image capture device is rendered insensitive to light.
8. A method as claimed in claim 4 or Claim 5 wherein the movement is continuous for the purpose of speeding up the scanning of a specimen.
9. A method as claimed in claim 8 wherein the continuous movement during the imaging results in blurring of the image, and the method includes the step of applying deconvolution to re-sharpen the image.
10. A method as claimed in any preceding claim wherein the excitation light is composed of light having two or more different wavelength(s).
11. A method as claimed in claim 10 wherein a single excitation light source is employed which comprises an acousto optic tuneable filter (AOTF) crystal and the wavelength of the emitted light is varied by altering the frequency controlling signal to the crystal as required.
12. A method as claimed in any preceding claim wherein the excitation light is pulsed.
13. A method as claimed in any preceding claim wherein the intensity of the incident excitation light is adjusted from one exposure to another by interposing neutral density filters, or opening or closing an iris diaphragm in the light path, adjusting the power to

the light source, or employing an attenuating element such as an AOTF or LCD shutter, or any combination thereof.

14. A method as claimed in any preceding claim wherein the specimen is illuminated by light at different wavelengths and the intensity is adjusted to produce a predetermined level of excitation intensity at the specimen for each wavelength.

15. A method as claimed in claim 14 wherein the adjustment produces a substantially similar level of intensity at the specimen for each different wavelength.

16. Apparatus by which light emitted from a specimen is imaged by an image capture device to produce a video signal for creating an image in a display device or for processing and analysis, comprising:-

- means for mounting the specimen,

- a light source for producing excitation light,

- a confocal scanning system adapted to direct excitation light in one direction towards, and thereby to scan an area of the specimen and also adapted to convey light emitted from the specimen as a consequence of the excitation light incident thereon, in another direction, which operates in use to scan typically repeatedly an area of interest of the specimen,

- an image capture device having discrete spatially distinct light sensitive regions on which light emitted from the specimen is focussed to form an image after being conveyed through the scanning system in the said other direction, and

- control means including a controller programmed to function as a state machine and adapted to control the scanning system, and excitation light and/or the image capture device so that light from the specimen is incident on the image capture device for a

specific time period equal to that required by the scanning system to scan the area of interest n times (where n is a whole number equal to or greater than 1).

17. Apparatus as claimed in claim 16 further comprising shutter means which in use is operated by signals from the control means to interrupt light from the excitation source except for when the specimen is to be illuminated wherein the shutter means comprises an acousto-optic element.
18. Apparatus as claimed in claim 17 further comprising second shutter means between the scanning system and the image capture device, which second shutter means is operated by signals from the control means so that in use light is prevented from reaching at least part of the image capture device sensor, except for the specific periods of time during which excitation light is incident on the specimen, for the purpose of reducing errors which could arise from phosphorescence, afterglow, stray reflections or light due to other effects, reaching the capture device.
19. Apparatus as claimed in any of claims 16 to 18 which further includes drive means adapted to move the specimen, the scanning system, or an element of an optical system within the scanning system, along a linear axis (the Z axis) so that in use the position of the plane can adjusted relative to the specimen.
20. Apparatus as claimed in claim 19 wherein in use the control system operates so as to restrict movement along the linear axis to periods during which light is prevented from reaching the image capture device.
21. Apparatus as claimed in claim 19 or claim 18 wherein the control system is operable to only produce movement along the linear axis during periods in which the excitation source light is inhibited or prevented from reaching the specimen.
22. Apparatus as claimed in claim 19 wherein the linear axis motion of the specimen, or scanning system, or element thereof, is continuous and wherein the apparatus further

comprises means by which deconvolution is applied to re-sharpen the image at the image capture device, or an image produced by signals from the image capture device, which is otherwise blurred due to the said continuous motion.

23. Apparatus as claimed in any of claims 16 to 22 wherein in use the wavelength of the excitation light is required to vary from one exposure to another, and the apparatus comprises two or more excitation light sources each producing excitation light of a different wavelength from the or each other source, and the control means is adapted in use to select the source to provide light of appropriate wavelength for each exposure.
24. Apparatus as claimed in any of claims 16 to 22 wherein in use the wavelength of the excitation light is required to vary from one exposure to another, the apparatus comprises a single source of excitation light which is adjustable to produce light of different wavelengths, and the control means is adapted to adjust the source to produce light having the required wavelength for each exposure.
25. Apparatus as claimed in any of claims 16 to 24 wherein the excitation light source is operable to produce light of more than one wavelength at the same time.
26. Apparatus as claimed in any of claims 16 to 25 wherein a single excitation light source is employed, the wavelength or wavelengths of the light emitted therefrom can be altered, and the control means is adapted to adjust the source to produce light of a desired wavelength or wavelengths wherein the light source is a laser light source which comprises an acousto-optical tuneable filter (AOTF) crystal, and the control means is adapted to provide signals to alter the frequency controlling signal to the crystal, to control the wavelength (or wavelengths) of the emitted light.
27. Apparatus as claimed in any of claims 16 to 26 wherein the excitation light source is operable so as to produce pulses of light.

28. Apparatus as claimed in any of claims 16 to 27 wherein the excitation light intensity is controlled by means of an attenuating element and the control means is adapted in use to control or position the attenuating element as appropriate.
29. Apparatus as claimed in claim 28 wherein the attenuating element is an AOTF or LCD shutter.
30. Apparatus as claimed in any of claims 16 to 29 wherein the control means is adapted to alter the intensity of the illumination so as to provide a predetermined intensity of illumination at the specimen for each wavelength, to remove variation in intensity from one wavelength to another as can occur due to inherent intensity variation as between one source and another or between different modes of operation of the excitation light source.
31. Apparatus as claimed in any of claims 16 to 30 wherein the control means is adapted to adjust the power to the excitation light source and/or control attenuation of light therefrom, from one exposure to another, to provide substantially constant intensity luminescence, to reduce variation in the intensity of the light incident on the image capture device sensor due to differing wavelengths of excitation light, or to render the light emitted due to luminescence of similar intensity irrespective of wavelength, or both.